Appl Microbiol Biotechnol (2013) 97:1829–1840
DOI 10.1007/s00253-013-4692-5

MINI-REVIEW

An overview of transducers as platform for the rapid detection of foodborne pathogens

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Received: 13 September 2012 / Revised: 29 December 2012 / Accepted: 2 January 2013 / Published online: 18 January 2013
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Abstract The driving advent of portable, integrated biosensing ways for pathogen detection methods offers increased sensitivity and specificity over traditional microbiological techniques. The miniaturization and automation of integrated detection systems present a significant advantage for rapid, portable detection of foodborne microbes. In this review, we have highlighted current developments and directions in foodborne pathogen detection systems. Recent progress in the biosensor protocols toward the detection of specific microbes has been elaborated in detail. It also includes strategies and challenges for the implementation of a portable platform toward rapid foodborne sensing systems.

Keywords Biosensor · Food · Detection · Pathogen · Transducer

Introduction

Food contaminated with pathogens is the natural cause of serious human health problems. Therefore, the detection of microorganisms in food is the only solution to secure human health and safety. Foodborne pathogens are the leading causes of gastrointestinal infectious diseases in developed countries. Along with the wide range of conventional methods for the detection of foodborne pathogens, polymerase chain reactions, enzyme-linked immunosorbent assays, and microarray-based techniques have also become prevalent in diagnostics. However, all these methods may take several hours or even a few days to conclude with some output of result. To overcome such factors, biosensors play a vital role in the rapid and efficient detection of foodborne pathogens. Moreover, the success of biosensor technology has diminished the large investments in basic biology facilities and individual research laboratories within a fraction of seconds (Bai et al. 2010). In addition, few more biomolecular techniques are under research pipeline to improve the selectivity and sensitivity of biosensors. Advancement in biosensors has greatly improved today's life by the detection of minute quantities of analytes within low detection limits. The detection of pathogens by these sensing techniques (Table 1) has been of immense interest in the food industry. Foodborne pathogenic biosensors can even detect trace amounts of microorganisms and toxins, which are responsible for the contamination of food. Biosensors are grouped according to the signal transductions they generate; it includes optical and electrochemical transducing systems. Optical biosensors measure the quenching, refractive index, fluorescence, chemiluminescence, and energy transfer and possess the advantage of rapid sensing. Electrochemical sensors are potentiometric or amperometric devices (Bhunia et al. 2010), very well addressed in our recently published article Arora et al. (2011). This article presents a review about the potential applications of both optical as well as electrochemical transducers according to their true potential in the detection of food pathogenic microbes such as *Escherichia coli*, *Salmonella*, and *Listeria*. It concludes that biosensors offer the most promising solutions and deal with some of the modern-day needs for the rapid and sensitive detection of foodborne pathogens in real-time or near-real-time devices.

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Biosensors for foodborne pathogen detection

The severity and frequency of outbreaks resulting from the intake of contaminated food has raised a significant concern in

Table 1 List of biosensor-based techniques commonly used for pathogen detection

Techniques	Pathogen detection
Optical biosensors	
1. Surface plasmon resonance	*Staphylococcus aureus* (Subramanian et al. 2006)
	E. coli O157:H7 (Waswa et al. 2007)
	Salmonella enterica (Koubova et al. 2001)
	V. cholera (Jyoung et al. 2006)
	Cryptosporidium parvum (Kang et al. 2006)
	Listeria monocytogenes (Leonard et al. 2004)
	Phytophthora infestans sporangia (Skottrup et al. 2007)
	Tobacco mosaic virus (Boltovets et al. 2004)
	Hepatitis A virus (Gomara et al. 2000)
	Salmonella typhmurium
2. Fibre-optic sensor	*Listeria monocytogenes* (Hamon et al. 2006; (Bierne et al. 2007; Ohk et al. 2010)
	E. coli O157:H7 (DeMarco and Lim 2002)
	Salmonella enteritidis
	Bacillus anthracis
	Staphylococcal enterotoxin B
3. Colorimetric and fluorescence detection	*E. coli* (Su et al. 2005)
	Salmonella enterica (Silbert et al. 2006)
	Bacillus cereus
Electrochemical biosensors	
1. Impedance-based field biosensor	*Salmonella* spp. (Louie et al. 1998)
	E. coli O157:H7
2. Electrochemical biosensor array	*Bacillus cereus* (Ertl and Mikkelsen 2001)
	Staphylococcus aureus
	Proteus vulgaris
	Escherichia coli
	Enterobacter aerogenes
	Saccharomyces cerevisiae
3. Impedance-based HRP-labeled immunosensor Ag-PSA-based DNA sensors	Rat IgG, HBsAg, HBeAg (Yu et al. 2006)
	Stachybotrys chartarum
	Escherichia coli
4. IDA-AP amplification-based RNA microarray sensors	*Escherichia coli* (Elsholz et al. 2006)
	Pseudomonas aeruginosa
	Enterococcus faecalis
	Staphylococcus aureus
	Staphylococcus epidermidis
5. IDA-AP amplification-based DNA microarray sensors	*Bacillus anthracis* (Liu et al. 2008)
	Yersinia pestis
	Francisella tularensis
	Ortho pox viruses
6. IDA-β-Gal amplification-based DNA microarray sensors	*Bacillus cereus* (Elsholz et al. 2009)
7. SPE-AP amplification-based array DNA sensors	*Listeria monocytogenes* toxin InlA (Laschi et al. 2006)
	Salmonella spp. (Farabullini et al. 2007)
	Escherichia coli O157:H7
	Staphylococcus aureus
8. HRP amplification-based DNA microarray sensor	*Bacillus anthracis* (Ghindilis et al. 2007)
	Yersinia pestis
	Escherichia coli

Table 1 (continued)

Techniques	Pathogen detection
	Bacillus subtilis
	Bordetella pertussis (Lodes et al. 2007)
	Streptococcus pyogenes
	Chlamydia pneumoniae
	Mycoplasma pneumonia
9. Hoechst 33258-based DNA array sensor	*Clostridium piliforme* (Goto et al. 2007)
	Helicobacter bilis
	Helicobacter hepaticus
	mouse hepatitis virus
10. HRP amplification-based DNA multiwell sensor strips	*Escherichia coli* O157:H7 (LaGier et al. 2007)
	Salmonella spp.
	Campylobacter jejuni
	Staphylococcus aureus
11. Esterase 2 amplification-based DNA array sensor	*Escherichia coli* (Pöhlman et al. 2009)
	Bacillus subtilis
	Bacillus atrophaeus
	Listeria innocua

the scientific world toward the discovery of fast, accurate, and ultrasensitive methods for the detection of foodborne infectious microorganisms. Contamination of food with pathogenic agents has become a matter of global concern due to its economic and social impacts. Hygienic conditions should be maintained during food production and packaging to avoid pathological outbreaks resulting from food ingestion. Therefore, an urgent need in the improvement of foodborne pathogen detection methods should considerably be highlighted to ensure safe and secure life. Previously, culture-based methods were in practice, which involve multiple steps and consume a lot of time to reach at the detection level. Even rapid methods of pathogen detection involving immunology-based methods and polymerase chain reaction also consume hours of arriving at some inference (Velusamy et al. 2010). The urgent need of quick and highly sensitive methods for pathogenic organism detection in food has led to the discovery of biosensors. These instruments met the needs of sensitivity, quick detection of foodborne pathogens, and low-cost analysis. Based on the method of signal transduction, biosensors (Fig. 1) can be optical, mass, electrochemical, magnetic, micromechanical, and thermal (Arora et al. 2011). Different types of biosensors work on different principles for analysis methods. The sensitivity of biosensor systems depends on the transducer's properties and on the various biological elements used for analysis (Palchetti and Mascini 2008). Fourier transform infrared spectroscopy, light scattering, surface plasmon resonance (SPR), fiber-optic, microfluidic protein biochip, and mammalian cell-based sensors were capable of effectively and efficiently detecting the presence of pathogenic microorganisms in food samples. Antibodies, nucleic acids, aptamers, bacteriophages, peptide nucleic acid, and locked nucleic acid act as biorecognition elements for pathogen detection in biosensor applications. Certain biosensors make use of labeled probes or reagents for the detection (Bhunia et al. 2010) of foodborne microbes. The future of biosensor technology in detection can be forecasted on the basis of the suitability of these sensors to get integrated in automated assays and the ability to perform in situ real-time analysis. The ideal sensor must have the potential of analyzing unknown samples in minutes and also must be capable of analyzing multiple agents simultaneously.

Aptamer-based biosensor recognition elements

The biorecognition principle classifies biosensors into immunochemical, receptor-mediated enzymatic/non-enzymatic, whole cell, and nucleic acid sensor systems. The biorecognition elements are used for analyte binding in biosensors. The sensitivity and selectivity of recombinant DNA technology alters the existing properties of many biorecognition molecules with few additional functions (Hock et al. 2002). The current generation of novel biosensing materials majorly relies on genetically engineered receptor molecules and genetically transformed cells. Biosensors using aptamer-specific nucleic acid sequences as biorecognition elements, which can suitably adhere to non-nucleic acid surfaces, are called aptasensors. These molecules are nucleic acid ligands such as RNA, ssDNA, modified ssDNA, or modified RNA (Fig. 2) that are identified and selected through an in vitro process called systematic evolution of ligands by exponential enrichment (SELEX), as reported by Torres-Chavolla and Alocilja

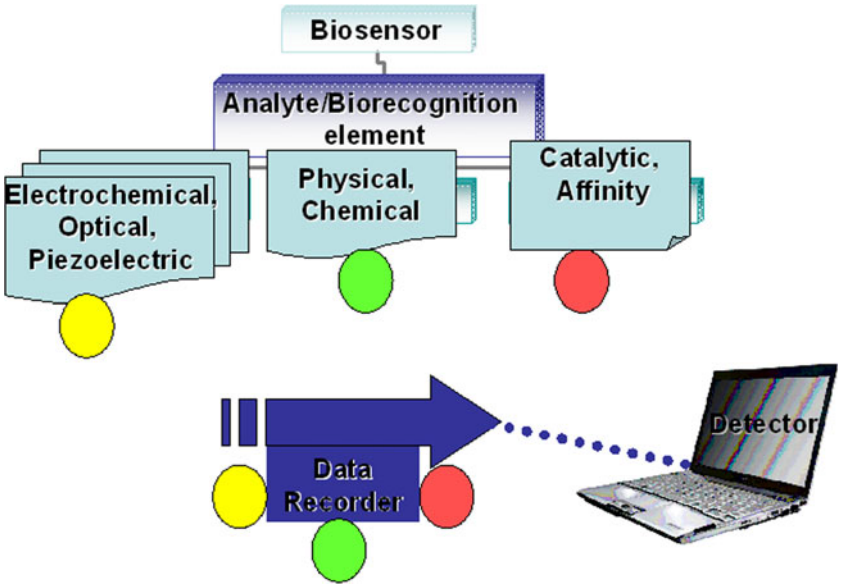

Fig. 1 Biosensor configuration for potential foodborne pathogen detection via analytical signaling

(2009), Ellington and Szostak (1990), and Tuerk and Gold (1990). Aptamers are artificial nucleic acid ligands generated by an iterative process of adsorption, recovery, and reamplification against amino acids, drugs, proteins, and other molecules (O'Sullivan and Guilbault 1999). Aptamers are mainly amorphous in solution; however, the association of aptamer–ligand folds them into several molecular architectures in which the ligand acts as an intrinsic part of the nucleic acid structure. The short, single-stranded, or modified oligonucleotides with self-annealing tendency help them to fold easily into 3D structural forms. Aptamer–target molecule association and adhesion on extensive scale have proven the complex suitable for analytic and diagnostic applications (Luzi et al. 2003). Proteins, whole cells, amino acids, oligosaccharides, and small organic molecules are the most common targets of aptamer selection, making them suitable as potential representatives for detection purposes. In emerging diagnostic and detection assays, aptamer recognition elements

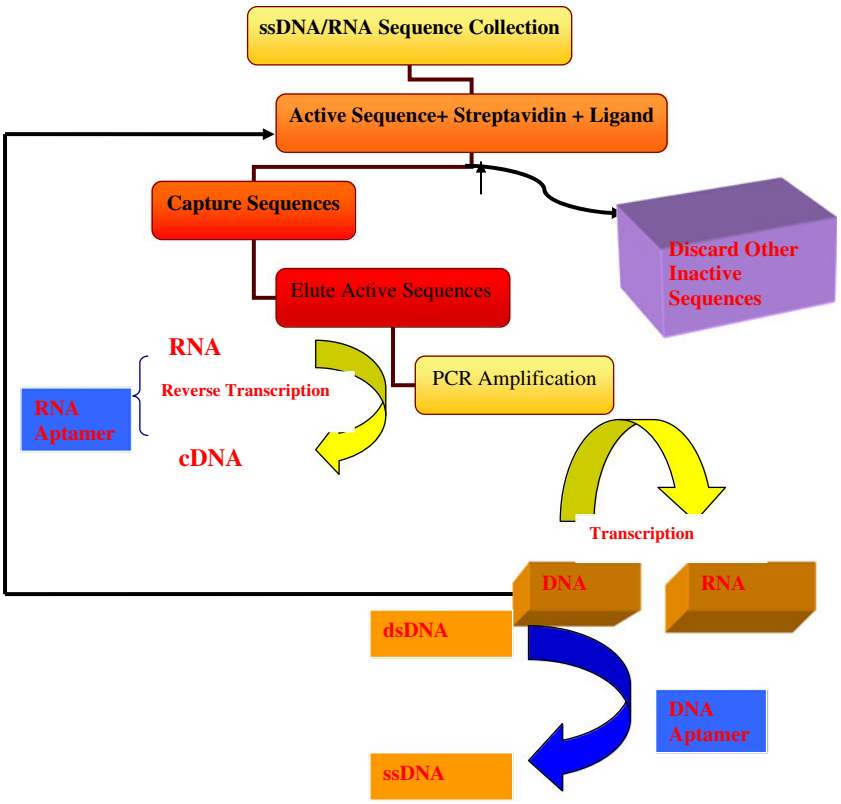

Fig. 2 Schematic flowchart for the isolation of aptamers (DNA/RNA) by SELEX. Initially, from the large library of small molecules, viz. DNA/RNA are selected and then targeted incubation leads to specific binding and partitioning of library via the elution of binding sequences from the target for the subsequent stringent selection of aptamers

represent an alternative to antibodies due to their high affinity, simplicity, and specificity. As key contributors toward DNA aptamer-based biochip, Stadtherr and co-workers have demonstrated the feasibility of protein detection (IgE and anti-IgE Ys; Stadtherr et al. 2005). Aptamer-functionalized cantilever surfaces have been successfully reported by Savran et al. (2004). Cellular detection is one of the promising areas of aptamer research. It possesses the key ability to target and discriminate microbial strains without knowing the membrane molecules or structural conformation in the cell. One such recent paradigm is capillary electrophoresis-based immunoassay for the evaluation of aptamer-based binding to bacterial cells. The aptamer affinity probe, capillary electrophoretic approach was conventionally proven through binding affinity studies. The aptamer probe was targeted with *Campylobacter jejuni* against other common foodborne pathogens, such as *E. coli* O157:H7 and *Salmonella typhimurium*, for specific detection. Moreover, this screening method does not require immobilization of the probe or target on a matrix, thereby aiding in reducing interference with conformational and binding strategies (Stratis-Cullum et al. 2009). In conclusion, the specificity exhibited for the probe bacterial target rationalizes an additional investigation for the qualitative screening of aptamer candidates for potential applications to food pathogen sensing. Furthermore, the significance of aptamers has been revealed on numerous biosensing platforms. A quartz crystal biosensor based on an RNA aptamer has been designed to detect HIV-1 Tat protein for the transactivation of the transcription process (Minunni et al. 2004). Interestingly, surface acoustic wave biosensor as an anti-thrombin aptamer was developed recently for monitoring blood coagulation cascade compound module (Gronewold et al. 2005). Toward this platform, now, first-generation magnoresistive biorecognition element-based spin valve sensors are fabricated to detect biological warfare terrorists such as *Bacillus anthracis*, *Yersinia pestis*, *Brucella suis*, *Francisella tularensis*, *Vibrio cholerae*, *Clostridium botulinum*, *C. jejuni*, and *Vaccinia* viruses (Graham et al. 2004). Recently, the sensor has been successfully developed using an optical microscope and RNA aptamers from *E. coli* O157:H7 (lipopolysaccharide), *Staphylococcus aureus* (teichoic acid), and *S. typhimurium* (membrane protein OmpC). The RNA aptamers were hybridized with thiol-conjugated 16 dT-linked molecules on a silver surface fabricated glass slide for the detection of foodborne bacteria via site-specific tagging/binding. The sensor system rapidly monitors individual food pathogens in a fraction of minutes (Maeng et al. 2012). Nature's effectiveness indicates that there is substantial untapped potential in small-molecule aptamers. On the contrary, very few researchers have chosen to invest the time and expense to develop aptamers from small molecules. As SELEX is time-consuming and demands labor-intensive efforts, still less than 30 % of selections result in aptamers (Gold et al. 2010). Additionally, the unique technical challenges imposed on the development of aptamers when selecting for small molecules have resulted in the success of very few novel aptamers that can bind to practical small molecular targets. Nevertheless, now, there are many opportunities available for the innovative applications of the role of small-molecule binding aptamers in biological and chemical biosensing systems to monitor contamination in our environment and food supply. The growing recognition is a clear stimulus to precede aptamer-based biosensing past the few proof-of-concept systems and to authenticate aptamer-based small-molecule sensing approaches for challenging the real-world bioanalytical innovative applications (McKeague and DeRosa 2012). There is still a requirement of continued effort in the development of small-molecule aptamers for foodborne microbes and other relative sectors to realize its true potential at its fullest.

Detection of *E. coli*

E. coli are found in the mammalian gut in symbiotic association with the host. A pathogenic strain of this bacterium generated due to mutations is capable of causing several human diseases such as traveller's diarrhea, infant diarrhea, and infections of the urinary tract. The mere presence of *E. coli* in food signals that food is unfit for consumption; therefore, food industries are mainly concentrating on preventing *E. coli* contamination in foodstuffs during manufacturing to packaging and until it reaches the hands of end users, "the consumers." Recently, silver nanoparticles were activated by biosorption for capturing this pathogenic bacterium. The biosorption of polyclonal antibodies (anti-*E. coli*) onto nanospheres and nanorice was done through a protein-A layer. Raman scattering surface enhancement confirmed the detection and was found promising due to the dimensions possessed by these nanodetectors. Just in 1 h, these nanomaterials had detected 10^3 cells/mL from milk and apple juice without any new enrichment. The technique could be conveniently used for detecting other pathogens using their pertinent antibodies (Naja et al. 2010). In 1982, a new strain of *E. coli* was discovered to cause severe hemolytic–uremic syndrome in young and elderly victims (20 %) leading to kidney failure (Rangel et al. 2005; Herold et al. 2004). This new serotype *E. coli* O157:H7, with Shiga toxin-producing genes, was believed to have been first transferred via bacteriophage (Herold et al. 2004). The Center for Disease Control found *E. coli* prevalent for 73,500 infections and 61 deaths/annum only in the USA. The prime source of *E. coli* infection is undercooked or raw beef during meat processing (Rangel et al. 2005). Conventional enrichment and plating methods require at least a day for cultures to grow. In such a situation, biosensors have an edge due to their rapid detection tests for food-processed pathogens. A new, rapid mechanical detection system with piezoelectric-induced resonance which

relies on the binding of *E. coli* to an antibody is extensively studied by Zuehlke (2007) and showed promiscuous for the future. Chen et al. (2008) reported a circulating-flow piezoelectric biosensor based on gold nanoparticle amplification for the real-time instant detection of a foodborne pathogen, *E. coli* O157:H7. A 30-mer thiolated probe-1 specific to *E. coli* O157:H7 *eaeA* gene was surface-immobilized and subsequently hybridized with the *E. coli* O157:H7 *eaeA* 104-bp amplified gene fragment. A second sequence complementary thiolated probe 2 was conjugated with the gold nanoparticles. It resulted in a mass change and consequent frequency shift during amplification to vary the target probe sequence of the piezoelectric biosensor. In *E. coli* O157:H7, the PCR products amplified within the concentration of 1.2×10^2 cfu/mL were recorded by the piezoelectric biosensors. The main advantages of the piezoelectric sensors are their easy adaptability, faster detection, and simplicity. In future, the piezoelectric biosensor may able to detect pathogens directly from raw food samples. For the rapid detection of foodborne *E. coli* O157:H7A pathogen, an electrochemical sandwich immunoassay biosensor (polyaniline-based) was developed. The biosensor is composed of a capture protein and a reporter protein. The reporter protein binds the target protein and the capture protein forms a sandwich complex. The two electrodes sandwiched the capture protein on a fixed pad, while the reporter protein adheres to the conductive polymers. The conductive polymer sheds electrical signal as a messenger, reporting the quantitative screening of the target protein. The architecture of the biosensor shows that it could detect about 7.8×10^1 cfu/mL of *E. coli* O157:H7 in 10 min (Muhammad-Tahir and Alocilja 2003). A label-free electrochemical impedance immunosensor on the principle of Faradic impedance spectroscopy for the rapid detection of *E. coli* O157:H7 was developed using an interdigitated array microelectrode (IDA). It immobilizes anti-*E. coli* antibodies onto an indium–tin oxide IDA. The electric model was proposed for understanding stepwise changes in an electrochemical cell and the IDA microelectrode system-based sensor circuit. The components of the immunosensor system (Chen et al. 2012) consist of an ohmic resistor (electrolyte between two electrodes), a double-layer capacitor (electron transfer resistor), and a Warburg impedance (around each electrode) for the interpretation of tests. The immobilized antibodies were characterized by cyclic voltammetry and measured by electrochemical impedance spectroscopy with $[Fe(CN)_6]^{3-/4-}$ as a redox probe. The linear changes observed in the electron transfer resistance were interrelated with the logarithmic concentration of *E. coli* cells, which ranges from 4.36×10^5 to 4.36×10^8 cfu/mL, with the detection limit of 10^6 cfu/mL in the sensor system (Yang et al. 2004). The electrochemical impedance immunosensor technique easily detects pathogenic bacteria by immobilizing specific antibodies onto the IDA successfully. A quartz crystal microbalance (QCM)-based DNA sensing dramatically amplifies *E. coli* O157:H7 via frequency shifts of 163 Hz. The nanoparticle bioconjugates with the QCM surface resulted in effective mass amplification. It is a rapid and cost-effective promising approach for the real-time detection of *E. coli* O157:H7 via a circulating-flow system (Wu et al. 2007).

Detection of *Salmonella*

An American scientist named Salmon discovered *Salmonella*. It has caused gastrointestinal illness in humans for over 100 years. *Salmonella* is a motile, Gram-negative, non-spore-forming, rod-shaped microscopic living creature. They are widely distributed in nature. Their primary reservoir is the intestinal tract of humans and animals. They spread through the feces of humans and animals to other people and the environment through cross-contamination. The ingestion of contaminated food such as beef, poultry, milk, eggs, or vegetables causes salmonellosis. Symptoms of salmonellosis include diarrhea, fever, and abdominal cramps from 12 to 72 h after the ingestion of contaminated foods and lasts until 4–7 days. The occurrence of different types of *Salmonella* varies; however, *S. typhimurium* and *Salmonella enteritidis* are the two most commonly found foodborne pathogens. At times, people infected with *Salmonella* may develop Reiter's syndrome in later stages, which includes frequent pains in joints, irritation of the eyes, and painful urination, leading to chronic arthritis. Rapid prognosis at the initial stages can hinder its severity from infants to elder people. Biosensor-based detection is a robust and promising boon for patients infected with foodborne salmonellosis.

S. typhimurium

Bacteria accounts for 91 % of total food contamination (Potter et al. 1997; Beran et al. 1991). *Salmonella* spp. contaminating different food products such as pasteurized milk (Ackers et al. 2000; Ryan et al. 1987; Dalton et al. 1997), eggs (Jones et al. 1995; Schroeder et al. 2005), chicken (Layton et al. 1997; Mahony et al. 1990; Snoeyenbos et al. 1969; Bokanyi et al. 1990), and cheddar cheese (Fontaine et al. 1980) are globally well acquainted. The magnetoelastic sensors are handy and do not require direct electrical contacts for interpretation; this makes them suitable for the remote detection of foodborne pathogens. Magnetoelastic materials are an unstructured combination of iron, nickel, molybdenum, and boron ferromagnetic alloys. Magnetoelastic sensors were immobilized with polyclonal antibodies for the detection of *S. typhimurium* in food products (Guntupalli et al. 2007). The use of this biosensor proved highly advantageous as it works on remote query phenomenon and enables the detection of bacterial species

in sealed and opaque containers. The binding of *Salmonella* to the antibody immobilized on the sensor surface changed the resonance parameters that were quantified by the shift in the sensor's resonant frequency. The three different food samples were observed in a detection limit of 5×10^3 cfu/mL, with a $2\times0.4\times0.015$-mm sized sensor, within a response time of about 2 min. Visual characterization studies confirmed that the measured change in frequency shifts was due to the adherence of *S. typhimurium* on the sensor surface. Hence, the sensor serves the purpose for the detection of readily available food products. To add to the research, Huang et al. (2009) documented multiple phage-based magnetoelastic biosensors (MEB) for the simultaneous detection of different biological pathogens sequentially from the same host. The biosensor was fabricated with the help of blocking (BSA) and immobilizing specific phage onto the magnetoelastic resonator's surface. In the detection system, a reference sensor was used as a control, while an E2 phage-coated sensor specific to *S. typhimurium* and a JRB7 phage-coated sensor specific to *B. anthracis* spores were considered for sensing from analytes. The biosensor coated with the specific phage responded by capturing cells/spores only on analogous and specific single pathogenic solution. The control and blocking agent alleviates non-specific binding. The results demonstrated that in the MEB, a number of bacterial spores were directly proportional to the masses of the sensor and inversely proportional to the sensor's resonance frequency. The MEB is the budding attempt toward the simultaneous monitoring of target analytes to detect foodborne pathogenic species with good selectivity. On the principle of a piezoelectric biosensor, a flow injection analysis (FIA) system was developed for the detection of *S. typhimurium*. The anti-*Salmonella* species-specific antibody was immobilized onto the surface of a quartz crystal with gold electrode coated through a polyethylenimine–glutaraldehyde (PEG) adhesive protocol and dithiobis-succinimidyl propionate (DSP) mechanism of coupling reaction. The PEG technique proved more significant than the DSP coupling. The biosensor had responses of 23–7 Hz in 25 min for *S. typhimurium* concentrations from 5.3×10^5 to 1.2×10^9 cfu/mL. Thus, it suitably proved the successful application of the FIA system (Jianming et al. 2006). Recently, the electrochemical biosensor approach with a total area of 3.2 cm and only 100 L of test sample was developed for the detection of foodborne pathogens. The three-electrode system uses printed circuit boards and a small AC excitation potential of magnitude 5 MV for detecting bacterial contamination. The small-sized device is fabricated on the principle of photolithography coupled with electrodeposition. A thin-film gold electrode coated with a biolinker binds the specific functionalized pathogen, which changes the electrical impedance, leading to confirm the presence of the pathogen onto the target. The best and low-cost sensor detects 500 cfu/mL concentrations of *S. typhimurium* within 6 min in the common preserved food.

Salmonella enterica

A major foodborne pathogen, *S. enterica* is of worldwide concern. In recent year's fruits, vegetables (DuPont 2007), almonds (Isaacs et al. 2005), and peanut butter (Centers for Disease and Prevention Peport 2009) are added over traditional meat and poultry sources of *S. enterica*. Continual antibiotic treatment of infection has led to the emergence of new multidrug-resistant *S. enterica* serovar *Typhimurium* DT104 (Centers for Disease and Prevention Report 1996). Since *Salmonella* is most commonly associated with food products, a sensitive and rapid detection method to assess food safety based on a biosensor approach was developed. Valadez et al. (2009) reported an evanescent wave fiber-optic assay to detect *Salmonella* in eggshell and chicken breast. On, a biotin–avidin optical fiber surface, an anti-*S. enteritidis* polyclonal antibody was immobilized to capture *Salmonella*. The anti-*S. enteritidis* Alexa Fluor 647-conjugated antibody (MAb2F-11) was used as the reporter. An evanescent wave from a laser at 635 nm excited the MAb2F-11. The fluorescence was detected by a laser spectrofluorometer at 710 nm. The *S. enteritidis*-specific biosensor detected its presence at 10^3 cfu/mL in pure culture and 10^4 cfu/mL with the egg and chicken breast samples. The results showed the completion of a reaction profile in <8 h. The assay presented in this study was cross-confirmed by the TRF system that further necessitates the equivalent enrichment time (2–6 h) and bacterial concentration. Thus, the fiber-optic system was found suitable for the detection of *S. enteritidis* from food samples in a shorter time period. Conventional microbiological techniques are require up to 7 days for the complete identification of the species, whereas stressed or injured cells still have the tendency to produce false negative results. The development of innovative biosensors such as antibody-immobilized microcantilevers can overcome the evident limitations of traditional techniques of being time-consuming, expensive, difficult to automate, sensitive, and the accuracy of quantitative methods. Lab-on-a-chip-based novel mass detectors are capable of online detection directly in a liquid medium without the need for any sample preparation. Portable biosensors were prepared by integrating biosensor to a microfluidic platform. They can detect the presence of pathogenic bacteria *S. enterica* serotype enteritidis in concentrations of 10^5 cfu/mL within 40 min. Mass sensitivity can be enhanced by fabricating and optimizing biosensors within a vacuum environment. A dip-and-dry technique enables detection at low concentrations of 10^3 cfu/mL, and due to the requirement of minute volumes, a biosensor working in a vacuum is capable of detecting even the presence of a single cell in the sample (Ricciardi et al. 2010).

Detection of *Listeria monocytogenes*

It is an opportunistic Gram-positive facultative anaerobic rod-shaped foodborne pathogen leading to deadly disease listeriosis that results in mild fever, diarrhea, nausea, or vomiting with a slight headache and will cause septicemia, meningitis, and meningoencephalitis in severe cases. Listeriosis can lead to spontaneous abortions and stillbirths (Doganay 2003). The severity of *Listeria* infections is due to its ability to flourish at very low temperatures; therefore, the US FDA has adopted a "zero-tolerance" policy for the presence of *Listeria* in food items. Leonard et al. (2004) had devised a new substractive inhibition assay using a BIAcore 3000 biosensor for the detection of *L. monocytogenes* with a polyclonal antibody. BIAcore is a commercial biosensor based on the principle of real-time detection. It detects the presence of pathogen in foodstuffs (Jönsson et al. 1991). It works on the basis of SPR (Quinn and Kennedy 1999; Scarano et al. 2010) and is used for the detection of whole cells (Fratamico et al. 1998; Quinn and Kennedy 2001), toxins (Daly et al. 2000; Rasooly 2001; Naimushin et al. 2002), illicit drugs (Dillon et al. 2003), as well as steroids (Fitzpatrick et al. 2003). The cells of the bacterium were first incubated with an antibody for binding and then the unbound antibodies were washed through centrifugation. Subsequently, the unbound free antibodies were passed through anti-F_{ab} ligand-coated sensor chip for efficient binding. The generated response, inversely proportional to the inhibiting cell concentrations, was able to detect 1×10^5 *L. monocytogenes* cells per milliliter in <30 min with minimum sample handling and preparation (Leonard et al. 2004). Currently, an antibody/aptamer-functionalized fiber-optic biosensor has also been developed for the rapid detection of *L. monocytogenes* from food products using a surface protein internalin-A (InlA) reporter involved in pathogenic infection (Ohk et al. 2010).

Detection of *C. jejuni*

The latest reported aptamer approach has recently gained success in the targeted detection of heat-killed and live *C. jejuni*. Live as well as heat-killed *C. jejuni* bacteria were added to the aptamer particles in equal ratios, and the induced magnet aids in the surface adherence of the desired probes for the targeted detection (Karkkainen et al. 2011). Additionally, a sandwich assay was also developed using aptamer-functionalized magnetic beads and quantum dots for the rapid detection of *C. jejuni* from buffers and different food products such as beef extract, chicken juice, and milk (Bruno et al. 2009; Karkkainen et al. 2011).

Detection of noroviruses

One of the major worldwide foodborne virus outbreaks of calicivirus includes norovirus (an RNA virus). It attacks specifically as a human pathogen and is responsible for most cases of viral epidemic gastroenteritis (Palchetti and Mascini, 2008). To detect the presence of noroviruses in the body, a team of scientists has identified the primary innate immune sensor. The sensor is based on a protein called melanoma differentiation-associated protein-5 (MDA-5). It stimulates an immune response that increases the body's defences to fight off murine norovirus-1 infection. The primary cellular sensor, MDA-5, may help develop a treatment that prevents or reduces norovirus infection. Toll-like receptor 3 is another such protein sensor which triggers the body's ability to detect the virus via recognizing dsRNA, and still more are yet to be discovered to detect the viruses for the prevention and treatment of human diseases (McCartney et al. 2008).

Biosensors for enterotoxin detection

Now, the biosensors are devised for the direct detection of toxins released by pathogenic organisms in food. These enterotoxins are responsible for the infection caused by the foodborne pathogens. *Staphylococcus* is one amongst the major microorganisms responsible for food poisoning. Infection by *Staphylococcus* has led to gastrointestinal diseases. *Staphylococcus* enterotoxins were resistant against the degrading effects of gastrointestinal enzymes as well as heat. For the detection of *Staphylococcal* enterotoxin B (SEB) released by *Staphylococcus* in food, an array biosensor was developed for analyzing multiple complex samples simultaneously. An array biosensor consumes less time in the detection due to its small dimension portability and facility for minimal sample preparation. Different food samples spiked with SEB are first incubated for 2 h; on the basis of fluorescent intensity, the detection of the toxin is confirmed. It was proven as a convenient assay for food safety validation. It can detect 0.5 ng of SEB per milliliter of sample in <20 min (Shriver-Lake et al. 2003), showing its smarter efficiency.

Piezoelectric immunosensors using gold electrodes were also used for SEB detection. An anti-SEB antibody was immobilized in different ways on the electrode. The best results in terms of sensitivity were obtained from an electrode coated with polyethyleneimine when analyzed in spiked milk samples. These biosensors proved highly specific for SEB detection as *Staphylococcal* enterotoxin D was not at all detected while *Staphylococcal* enterotoxin A (SEA) at a concentration of 40 μg/mL gave 6.44 % of the signals. The major concern with a piezoelectric biosensor is the stability of its crystal. It must be stored at 4 °C in a dry atmosphere for

preserving its activity and can be used for 3 days (Lin and Tsai 2003), showing its success in the detection.

The detection of *Staphylococcal* enterotoxin A can also be carried out using self-assembled monolayer screen-printed electrodes. The electrodes were coated with antibodies specific for the SEA. Firstly, protein A was immobilized on the electrode followed by an anti-*Staphylococcal* enterotoxin antibody (IgG), then enterotoxin A, and an anti-*Staphylococcal* enterotoxin antibody conjugated with peroxidase was coated with each immobilization step for 1 h. These biosensors showed promising results for *Staphylococcal* enterotoxin detection in phosphate buffer (Pimenta et al. 2010). SPR biosensors are also capable of detecting *S. aureus* enterotoxin B. A sandwich assay is devised through the immobilization of SEB from standard solutions or samples. Furthermore, optimization was carried out in HEPES buffer and in ham extracts. The minimum detection limit of this assay is capable of detecting 2.5 ng/mL of bacterial in HBST buffer or in the ham tissue extract. For improving the sensitivity, a more specific antibody can be induced in the process. The Biacore method is semi-automated and can be developed for analyzing multi-toxins in food samples (Medina 2003), at a glance.

C. botulinum produces neurotoxins that are responsible for causing flaccid muscular paralysis in the affected person. *Clostridium* releases neurotoxin types A, B, C, D, E, F, and G. A fiber-optic-based biosensor has been devised for the detection of *C. botulinum* toxin in food samples. The evanescent wave of a tapered optical fiber is used for signal discrimination that had made this method rapid and very sensitive. A 50-mW argon ion laser is used as a source for laser light generation. The optical fiber probe is tapered at distal ends, and rhodamine-labeled polyclonal antitoxin A immunoglobin G (IgG) antibodies were covalently attached to it to generate a good sensor. Different fluorescent signals were generated as a result of antibody and toxin conjugation. A number of antitoxin antibodies can be immobilized to the fibers. Using this biosensor, botulinum toxin A can be detected even at low concentrations of 5 ng/mL within a minute. Affinity-purified polyclonal horse antitoxin A antibodies showed better results than an IgG fraction obtained from the same horse serum or monoclonal antitoxin A antibody BA11-3. The reaction between the antibody and botulinum toxin A shows high specificity in the assay. As yet, no response was reported against tetanus toxin (Ogert et al. 1992), showing the potential authenticity of the sensor.

Conclusion

In order to meet recent outbreaks, the resulting fatalities, and economic losses due to foodborne pathogens, significant attention toward the rapid detection of these pathogens moves in the veins of researchers. There is a vital necessity for rapid, efficient, and reliable methods for the detection and identification of pathogens to comply with the demands of consumers for safer food. One such way is the microspot, a color change paper-based test system which has been demonstrated for the detection of selected foodborne pathogens such as *L. monocytogenes*, *S. typhimurium*, and *E. coli* O157:H7. It was capable of recording the index in concentrations as low as 101 cfu/cm^2 in 8–12 h from ready-to-eat meat samples (Jokerst et al. 2012). The rapid testing system indeed is a requisite demand, which gives reliable results in a shorter time with the help of biosensitive elements (biorecognition elements). In this review, we have highlighted new sensing technologies for specific foodborne pathogen detection that have taken advantage of advanced micro- and nanotechnology. Toward this end, transducers are the powerful analytical tools for diagnosis, particularly when quick response, low cost, simplicity, high sensitivity, and the inherent specificity of measurements are necessary for the detection of foodborne pathogens. However, the challenge still remains to make them off-the-shelf devices available for long-term routine use for the ease of common people monitoring at glimpse.

Acknowledgments First author gratefully acknowledges the Council of Scientific and Industrial Research (CSIR), Government of India, for providing the Senior Research Fellowship.

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